

Microcystin Monitoring at Metropolitan Water District

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History of Toxin Monitoring at Metropolitan

- AWWARF study on microcystin occurrence in 1995-1997 included MWDSC.
- Two major algal blooms in July 2001.
- Filtered algal bloom samples sent to Dr. Boyer's lab in Syracuse, NY.
- Algal toxin project initiated in May 2003.

Detection of Algal Toxins

- Bioassays: mouse, brine shrimp, rat hepatocytes
- ELISA
- Protein phosphatase inhibition (PPIA)
- HPLC/PDA (photodiode array)
- LC/MS
- Others (experimental or less often used)

Test Kits Used in Water Quality Lab

- Based on ELISA
- Made by Envirologix Inc., Portland, Maine
- Plate and tube kits differ in methodology, standards, analytical time
- Can detect microcystins and cross-react with nodularin.
- Can provide some quantitation & screening capability.
- Do not permit identification of toxins.

Comparison of Two Test Kit Methods for Microcystin

Plate

- LOD 0.147 ug/L
- Standards: 0.16, 0.6, 2.5 ug/L
- Time: about 2 hours
- Quantitative
- Requires plate reader
- Use: best for water samples
- Price: \$396

Tube

- LOD 0.3 ug/L
- Standards: 0.5, 3.0 ug/L
- Time: about 45 min.
- Semi-quantitative
- Read from color intensity
- Use: as screening tool
- Price: \$145

Principle Behind ELISA Tests for Microcystin

- Sample + antibody-coated surface (wells or tubes) → microcystin bound to antibody
- Enzyme-linked antigen + antibody → binding to remaining unoccupied sites
- Substrate + binding sites → color development inversely proportional to microcystin in sample

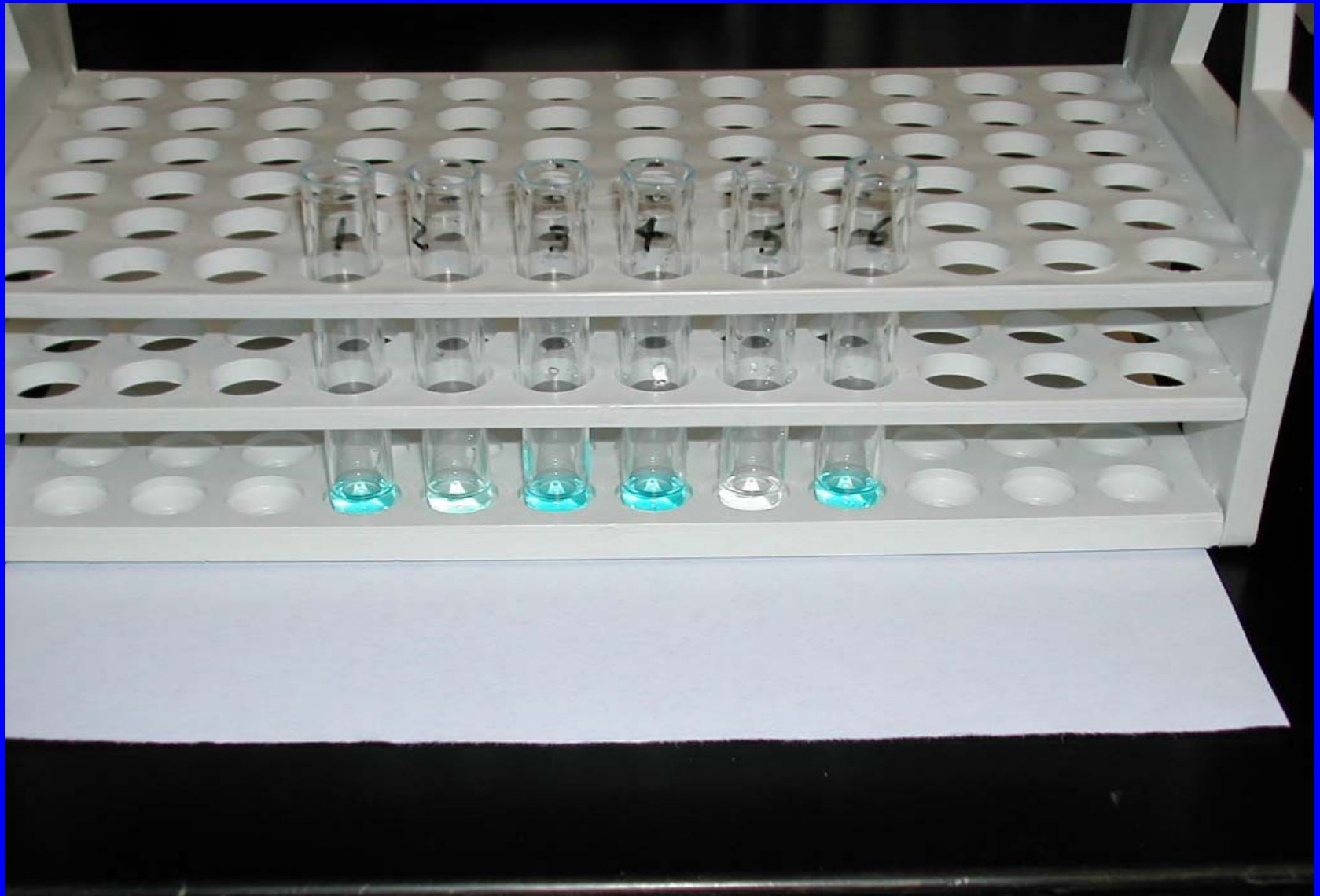
Microcystin Monitoring at MWD

- Monthly, all year.
- Lakes Castaic, Mathews, Perris, Skinner, Silverwood, Diamond Valley, & Calif. Aqueduct.
- Water samples and some benthic algal samples.

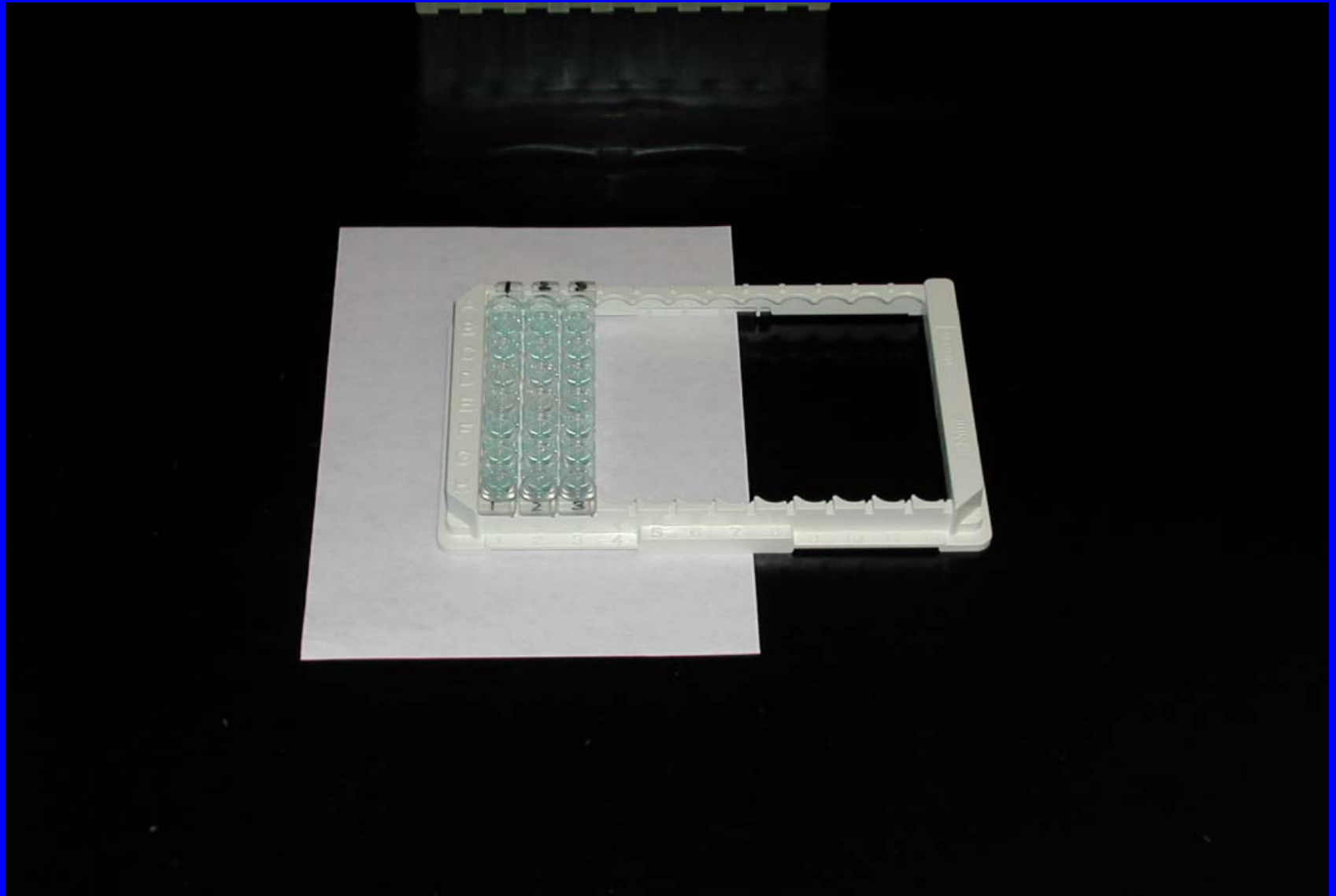
Microcystin Tube Kit and Accessories



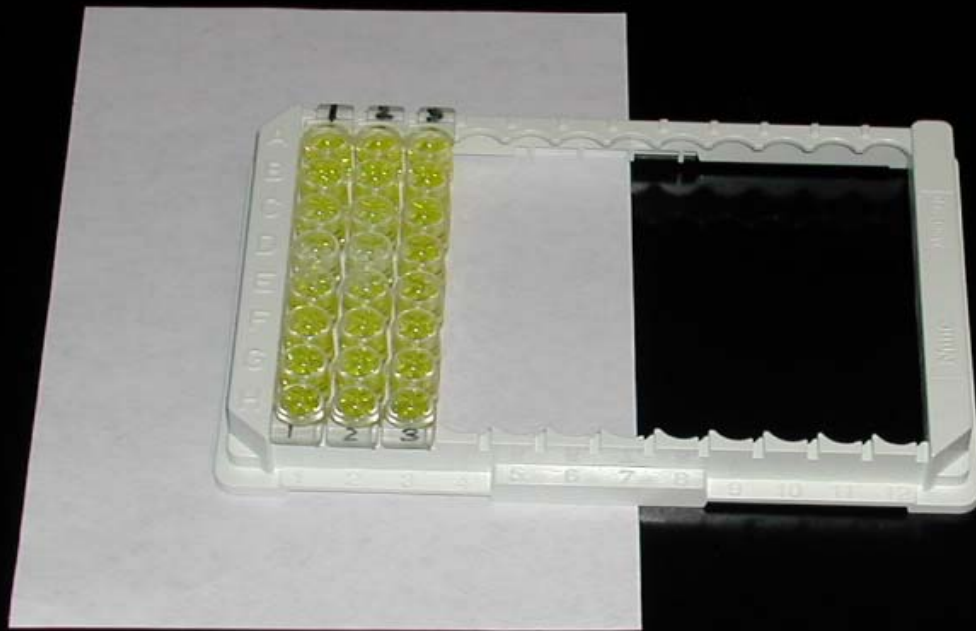
Completed Tube Assay



Wells in Plate Assay Showing Color Development



Wells in Plate Assay After Acidification



Tecan Plate Reader with Computer and Printer



Data Reduction Worksheet

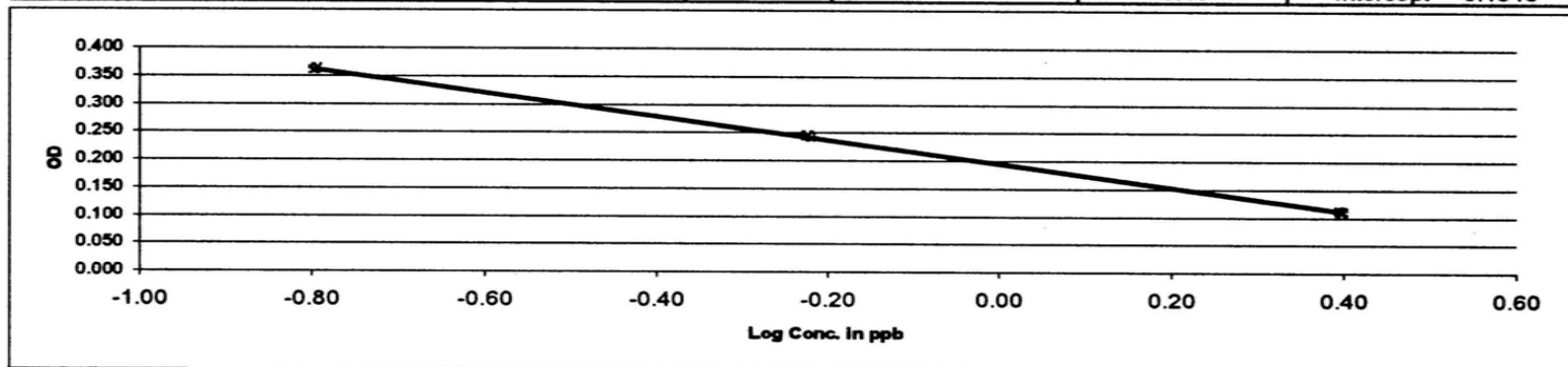
Operator:
Date:
Time:
Kit Lot#:

G. Izaguirre
9-Dec-05
11:15
82275

Section I) Calibration Curve

Calib. Conc.	rep1	rep 2	Abs. Value	%CV	B/Bo	Log(Conc.)	
Neg. Ctl. Abs. =	0.414	0.414	0.414				
0.16 ppb Abs. =	0.361	0.361	0.361		0.87	-0.80	
0.6 ppb Abs. =	0.260	0.226	0.243	9.9%	0.59	-0.22	
2.5 ppb Abs. =	0.105	0.114	0.110	5.8%	0.26	0.40	

$R^2 = -0.9999$
 Slope = -0.2107
 Intercept = 0.1943



Section II) Sample Calculations

Sample	Paste below	SAMPLE				Estimated PPB
	DATE	rep 1	rep 2	ABS	%CV	
1	14-Nov-05	0.40	0.39	0.395	2.3%	0.11
2	2-Nov-05	0.39	0.38	0.386	1.1%	0.12
3	9-Nov-05	0.19	0.17	0.175	8.1%	1.23
4	9-Nov-05	0.40	0.38	0.393	3.4%	0.11
5	30-Nov-05	0.41	0.43	0.416	3.7%	0.09
6	21-Nov-05	0.43	0.41	0.420	3.5%	0.09
7	5-Dec-05	0.40	0.38	0.387	3.5%	0.12
8	9-Dec-05	0.39	0.39	0.388	0.7%	0.12
9				#DIV/0!	#DIV/0!	#DIV/0!
10				#DIV/0!	#DIV/0!	#DIV/0!
11				#DIV/0!	#DIV/0!	#DIV/0!
12				#DIV/0!	#DIV/0!	#DIV/0!

LM

Summary of Cyanotoxin Results

- Water samples analyzed: 226
- Number positive for microcystin: 25
- Percent positive: 11.1%
- Most samples $<0.2 \mu\text{g/L}$
- Range: $<0.16 - \sim 15.7 \mu\text{g/L}$
- Samples $>1.0 \mu\text{g/L}$: 9 (4.1%)
- Detection limit by plate method (most sensitive): $0.147 \mu\text{g/L}$

Benthic Microcystin Producer

- Green *Phormidium* 10-14 µm in diameter.
- Found in Lakes Mathews, Skinner, DVL and Perris.
- Is very common component of benthic community.
- Has been analyzed in outside lab and toxin confirmed.

Benthic Microcystin Producer (Lake Mathews)



Recommended Reading

- “Everything a Manager Should Know About Algal Toxins but Was Afraid to Ask,” by Dr. Judy Westrick, *Jour. AWWA*, Sept. 2003.
- “Cyanobacterial Harmful Algal Blooms (CyanoHABs): Developing a Public Health Response,” by Lorraine C. Backer, *Lake and Reservoir Management*, March 2002.

Regulatory Issues

- USEPA Candidate Contaminant List (CCL) in 1998 included freshwater cyanobacteria and their toxins.
- USEPA workshop in 2001 identified 3 toxins for possible inclusion in CCL: microcystin, anatoxin-a and cylindrospermopsin.
- WHO guideline for microcystin-LR in drinking water (1998) set at 1.0 $\mu\text{g/L}$.
- Australian advisory limit for anatoxin-a: 3 $\mu\text{g/L}$.